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Knots in Macromolecular Systems: Concepts and Challenges

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The following survey of computational and experimental research activities on knots in synthetic and biological macromolecules outlines previous accomplishments and current challenges in the field.

1 Introduction

Knots are one of mankind's oldest and most practical devices. Appreciated by fisherman, seafarers, surgeons and rescue professionals around the world, knots have long captured the imagination of natural scientists, too. In 1867, Lord Kelvin suggested that atoms may consist of knots formed by the ether,[1] which inspired physicists like Maxwell and Tait to lay the foundations of knot theory.

In mathematics, knots are only well–defined in closed curves and usually categorized according to the minimal number of crossings in a projection onto a plane. The search for an algorithm which can distinguish between all knots is ongoing and still one of the grand challenges in mathematical knot theory. From a practical point of view, however, several algorithms, like the Alexander polynomial or the HOMFLY polynomial, are able to distinguish between simple knots and suffice for most purposes. In this spirit, open chains can also be analyzed by simply connecting end points in a defined way.

After descending into mathematical obscurity for some time, interest in the subject was revived in the 1960s, when Frisch, Wassermann[2] and Delbrück conjectured that all sufficiently long polymers have to contain knots - experimental length scales were, however, not known at this time. From a theoretical point of view, this statement is intriguing because knots are not included in the standard theory for self-avoiding polymers, but affect equilibrium properties and dynamics.

2 Computer Simulations of Knots in Polymers

Simulations are particularly well-suited to address these questions because the statistics of knots can be unveiled by analyzing independent polymer configurations generated by the computer. Starting with the ground-breaking work of Vologodskii et al.,[3] a variety of coarse-grained polymer models were tested.

With little or no excluded volume (random walk) and no inherent stiffness, a single three-dimensional polymer displays a large number of very small and localized knots. Each monomer can be positioned anywhere around its predecessor which promotes entanglements on the local scale. If excluded volume is considered, however, the chain becomes unknotted.[4] A single bead–spring polymer with 1000 monomers in good solvent

conditions, e.g, only contains knots in about 1% of all configurations.[5] The fraction of the polymer which is occupied by the knot in one of these rare events is considerably larger than in random walks, but still small with respect to the total chain–length. If a polymer collapses into a globular state and has enough time equilibrate, or if it is confined into a capsid, knots become frequent again[5] and spread out all over the globule.[5] In such a dense phase, the same bead–spring model contains knots in 80% of all configurations - most of which are already quite complex. In this case, the free energy of the polymer is dominated by energetic contributions which do not distinguish between unentangled and entangled states which are otherwise favored by conformational entropy. In the swollen phase, on the other hand, the polymer would like to have access to as many states as possible, and the free energy is dominated by entropic contributions which disfavor entanglements.

Analogies between microscopically knotted polymers and macroscopically knotted strings and robes are far reaching. Not only do single polymers and robes become knotted in crowded conditions, they also share similar material properties. A knotted robe, e.g., withstands only a fraction of the traction which it can withstand if the robe remains unknotted - a well-known fact among mountaineers and anglers. Knotted polymers are also far less stable under tension and tend to break at the entrance to the knot.[6]

3 Knots in DNA, Proteins and Synthetic Polymers

Knowledge gained from computer simulations is particularly relevant for biological systems whose fundamental constituents are long biopolymers. Knots were first discovered in bacterial DNA in 1976.[7] Since then, many knots were also created artificially, e.g., by the action of topoisomerase I on circular DNA. In good solvent conditions, DNA behaves like a model polymer and contains almost no knots (0.5-4% knots in a 10000 base pair strand depending on salt concentration.)[8] From the simulation of knotted homopolymers, one might expect that DNA also becomes highly knotted in confinement. This condition is, however, problematic for DNA in crowded environments like the cell, and indeed, nature has developed successful strategies to circumvent this effect. Human DNA, e.g., wraps around histone proteins. In ds-DNA viruses, the rather rigid DNA forms a spool when it is fed into the capsid (with one end remaining attached to the loading channel.)

Topology is even more relevant in proteins because the three-dimensional structure of a protein directly determines its functionality. Once again, knots are rare[9–12] although the reason is not well understood. It has been hypothesized that knotted structures are difficult to fold[11] and would essentially preserve their unknotted state after the initial collapse.[12] However, experiments suggest that certain knotted proteins may fold and unfold reversibly under a change of solvent conditions.[13] Secondary structure and inherent stiffness of the protein backbone may also simply shift the length scale at which knots occur in comparison to globular polymers.[12] Overall, the problem remains essentially unsolved. Interestingly, knots in proteins are usually preserved in structural homologs throughout evolution which indicates that they are relevant for the functionality of the protein and have existed since the beginning of life. In our recent comprehensive analysis of the Protein Data Bank,[9] we were also able to identify one counter-example and demonstrate how the presence of a knot may alter the enzymatic activity of the protein.

The synthesis of man–made molecules with knots has also been an important topic in the chemical literature.[14] These so-called “knotanes” consist of small knotted molecules obtained by supramolecular-template techniques.

4 Challenges

In the near future, I would expect that closed knotted nanoparticles based on knotted polymer globules will be synthesized.[15] These particles should be highly knotted, in the order of 10 nm in size and offer a reactive surface which may turn them into interesting candidates for the delivery of drugs. Similarly, it should become feasible to genetically alter proteins such that they become knotted. From a theoretical point of view, it would certainly be interesting to quantify why there are so few knots in proteins and how knotted proteins actually fold. 140 years after Kelvin, knotty problems persist.

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